ALGOLOGICAL NOTES

III. A Wood-penetrating Alga, Gomontia lignicola, n. sp.

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Some years ago while collecting on the island of Nashawena, one of the Elizabeth Islands near Woods Hole, Massachusetts, I found in a small fresh-water pond a yellow pine board which was more or less covered with a blue-green slime and accordingly it was taken to the laboratory for further investigation. The blue-greens proved to be nothing unusual, but occasionally there appeared irregular grass-green cells which seemed to be some unicellular form, and, since this could not be identified, the board was retained for further study. Investigation soon showed that the single cells were capable of germination, producing filaments of a millimeter or two in length, but it was not until the last few months, after the alga had been kept under observation for more than six years, that enough of the life history could be determined to identify the genus. Although from the first it was noted that the best way to obtain single cells was to scrape off the accumulation of *Oscillatoria* and *Aphanocapsa* on the surface, it was not recognized until later that the filaments were actually imbedded in the tissues of the wood and that this was not due to disintegration; in fact the pine board was in an excellent state of preservation, the part not submerged partially retaining its yellow color. As soon as the typical sporangia were secured it became evident that the alga was a *Gomontia*,—a fact which, from its habitat, might have been guessed before. But it must be confessed that in spite of this hint, I failed entirely to note the affinities of my plant with that of *Gomontia polyrhiza* until after the life history had been fairly well made out.

While the species to which I shall refer as *G. lignicola* has the well-marked characters of the genus, it differs in several distinct particulars from the species hitherto described, namely, the original one of Bornet and Flahault, *G. polyrhiza* (Lager.) B. & F.,[1] *G. codiolifera* (Chodat) Wille,[2] *G. arrhiza* Hariot,[3] *G. Holdenii* Collins,[4] and *G. Aegagropilae* Acton.[5] Chodat casually mentions another species, *G. Manxiana*,[6] but there is no description and no discussion which would enable one to judge the character of this plant. Miss Acton states that there is no valid reason for separating *Foreliella* from *Gomontia*, and *Foreliella perforans* Chodat should be known as *Gomontia perforans* (Chodat) Acton, so perhaps this species should likewise be added to the list.

Vegetative Condition

The filaments of *G. lignicola*, neither in their general appearance nor in their habit of growth, closely resemble the various species of the genus hitherto described. Except in young plants developing directly from zoospores (pl. 13, fig. 4) the individual cells are almost constantly cylindrical in form, from three to ten times longer than wide. Branching is so infrequent as to lead one to suppose the filament to be typically simple, although occasionally lateral branches of a few cells only may occur. The beginning of such a branch is shown in pl. 14, fig. 4. No indication of the rosette arrangement of branched filaments described for *G. polyrhiza* was observed, and the habit of growth precludes any such formation.

A most striking appearance is produced in the filament by the rather common condition of having the most of the chlorophyll concentrated in the terminal cell. A filament of a dozen or more cells, all approximately of the same size and shape, will, with the exception of the terminal cell, be so devoid of color as to give the appearance of a fungous hypha. Closer examination under a high-power objective reveals a thin thread-like chloroplast which runs for the length of the cell,

[1] Bornet, E., et Flahault, C. H. Soc. Bot. Fr. Bul. **36** : CLII. 1889.

[2] Chodat, R. Bul. de l'Herb. Boissier **6** : 443. 1898.

[3] Hariot, P. Soc. Bot. Fr. Bul. **38** : 417. 1891.

[4] Collins, F. S. Erythea **5** : 95. 1897.

[5] Acton, E. New Phytol. **15** : 102. 1916.

[6] Chodat, R. Algues vertes de la Suisse **1** : 61. 1902.

but the contrast between the brilliant bright green terminal cell and the pale, almost colorless, contiguous cells arrests the attention at once. If the spore from which the filament has arisen be still attached, as is frequently the case, it too may retain its brilliant green color, giving the effect of two algal cells being held together by a fungal thread (pl. 14, figs. 1, 4, 5). That this condition is perfectly normal and not due to any parasite or injury to the colorless cells is easily demonstrated by watching the formation of new cells as they are cut off apically from the original cell. It is as though the full supply of chloroplast material for the entire filament were contained in the apical cell and only enough supplied to the new cell to maintain it. The temptation to speculate upon the possible explanation of this phenomenon, which, together with the wood-penetrating habit, may be so easily construed as another indication of the origin of certain aquatic fungi from algae, is resisted in order that those who are more interested in this sort of thing may have the entire field to themselves.

The composition of the cell wall, as well as the cell contents, is typically Gomontia-like. No reaction for true cellulose is obtained either from the delicate wall of the young cells or the cutinized older and much thickened walls. From one to six rather large nuclei may be observed in each cell, and these do not seem to be, as suggested by West,[1] "due to the incipient formation of resting akinetes." A single chloroplast, which, because of its reticulate and irregular character, frequently gives the appearance of numerous disk-shaped or elongated color bodies, is present and a varying number of pyrenoids can usually be made out. Starch is very abundant in the resting cells, but may or may not be detected in the actively growing vegetative cells. Protoplasmic connections between the cells can easily be discerned, particularly in those cells practically devoid of chlorophyll (pl. 13, fig. 2).

Sporangia

Except for the absence of "rhizoids" the formation and general character of the sporangium of *G. lignicola* agree well

[1] Algae 1 : 305. 1916.

with the description of *G. polyrhiza.* Certainly, as Bornet and Thuret say, "Rien de plus variable que la configuration des sporanges." No two of the large number observed are of the same size or shape, and sporangia formed on the surface, a not infrequent occurrence, are just as irregular in outline as those produced in the tissue of the wood. The contents of the sporangium breaks up into zoospores simultaneously and not successively and the spores escape through a small hole invariably produced in the tip of one or more of the branch-like projections from the main body of the sporangium (pl. 14, figs. 2, 3). Bornet and Thuret were not able to observe zoospore discharge and thought possibly the entire sporangium broke up, thus liberating the spores. This is not the case in *G. lignicola,* however. A most satisfactory and simple means of obtaining discharging zoospore material is to transfer sporangia which have developed on unsubmerged parts of the wood. A part of the board which projected above the surface of the water but, because of the cover on the jar, was constantly in a moist atmosphere, was filled with sporangia. If these were removed to a drop of water on a slide, zoospores would almost immediately begin to be discharged, one at a time, through the minute opening. In fact, the best sporangium material was obtained in this way, and it would appear that a habitat permitting at least semi-aërial conditions is best suited for sporangium formation. So far as known, all the other species of *Gomontia* are to be found in situations which may permit either periodical or irregular intervals when the plant is not submerged, and it seems reasonable to suppose that such a habitat is conducive to the formation and discharge of zoospores.

Zoospores

In the account of the original species of *Gomontia,* Bornet and Flahault describe the zoospores as having two cilia of equal length and of two sizes,—a small one measuring 5×3.5 μ and a larger one $10–12\times5–6$ μ. No conjugation, either between small ones of the same size or between a large and small one, was observed. The direct germination of the large zoospore is, however, described.

Wille[1] definitely detected four cilia in zoospores of *G. polyrhiza*. The sporangia from which these came, however, can hardly be regarded as typical, since they were only about 15 μ in diameter and but from two to four zoospores were produced in a single sporangium.

The zoospores of no other species have been satisfactorily described and various authors have given the number of cilia for the genus as either two or four, according as to whether they followed Bornet and Flahault or Wille, or as in some instances regarding the biciliate spores as gametes and the quadriciliate as true zoospores.

All of the motile spores seen from the plants of *Gomontia lignicola* possess but two cilia (pl. 13, fig. 3). Because of the above described method for obtaining these spores an unusual opportunity was afforded to examine zoospores before, during, and after discharge. Thousands were seen and hundreds stained with weak iodine, which was sufficient to demonstrate the cilia. In no case was there the slightest indication of more than two cilia. Neither was there any marked difference in size as described for *G. polyrhiza*. The contents, however, of the zoospore of *G. lignicola* corresponds well with that of *G. polyrhiza,* having the single pyrenoid, a basal chloroplast, and the distinct red spot described for that species.

The possibility of *Gomontia* being one of the genera which possess a quadriciliate zoospore and a biciliate gamete has, of course, been recognized from the first. There is no account, however, of conjugation being seen, and the existence of gametes in *Gomontia* is based upon analogy rather than upon observation. So far as *G. lignicola* is concerned it can, I think, be regarded as highly improbable that gametes occur, at least it is certain that the biciliate bodies observed function as true zoospores. The fact that at least several hundred sporangia were from time to time observed discharging zoospores

[1] Über die Zoosporen von Gomontia polyrrhiza (Lagerh.) Born. et Flah. Norske Videnskab. Selsk. Skrif. **1906**[3]: 29. 1906.

and that the majority of these spores were followed through their germination, both in mixed cultures and under control conditions, justifies one in the belief that there is no foundation for the idea that simply because biciliate and quadriciliate spores have been recorded in *Gomontia* those possessing two cilia are gametes.

Germination of Zoospores

The germination of the zoospore may occur in one of two different ways, there being no visible distinction between those which produce a vegetative filament direct and those forming resting spores which in the past have erroneously been regarded as akinetes or aplanospores. The direct germination of a zoospore to produce vegetative cells was observed by Bornet and Flahault and regularly occurs in *G. lignicola* (pl. 13, fig. 4). Fully as many zoospores fail to germinate immediately, but after losing their cilia assume a spherical form and grow into a large irregular-shaped cell which ultimately produces, at from one to several points, vegetative filaments (pl. 14, figs. 1, 5). Although the method of forming the mature thallus is, of course, different, this body may roughly be likened to the polyhedral cell formed by the zoospores of *Hydrodictyon.* The zoospores which develop into resting spores, instead of germinating directly, retain the red spot for several weeks and the process of growth into a mature resting spore is a slow and gradual one. There is almost as much irregularity in the ultimate size and shape of the resting spore as there is in the sporangium. Plate 15 gives some idea of the great diversity in form of these spores but there is no limit to the variety which might have been shown, since scarcely any two are of the same outline. These spores were the cells first found by me in examining the scrapings from the pine board, and it must be confessed that even after their origin was discovered they were suspiciously reminiscent of certain unicellular grass-greens which have been described.

The resting spores derived from zoospores are brilliantly green in color and full of starch. A single pyrenoid is plainly visible in the young cells, but as they become more mature no pyrenoids can be observed. Simple staining methods showed but one nucleus. Later the cells became so packed with chlorophyll and starch that it was not possible without sectioning to determine satisfactorily whether or not this nucleus divided previous to the actual germination of the spore. The wall of the resting spore is always decidedly thickened at maturity and various lamellate excrescences and protuberances, referred to by other authors as "rhizoids," are not infrequently formed. The spore, after reaching mature size, may rest for months or even years. No change in nutrient solution or environment seemed to have the slightest effect on inducing germination, although almost every conceivable combination was tried. From time to time certain spores, either in pure or mixed cultures, would begin to germinate and there was no correlation between the results and the treatment to which the various cultures had been subjected.

The germination of the resting spore may apparently take place at any point and usually germ tubes push out at more than one place on the surface of the cell. Ordinarily these do not appear simultaneously but after a filament of several cells has been produced a new filament will start at some other point and this may be repeated until as many as four new filaments arise from a single spore. The resting spore is seemingly inexhaustible, retaining its bright color and abundant starch content until all the filaments produced are of considerable length. Then it slowly disintegrates, frequently leaving remnants of the old spore wall attached to the basal cell of the filament.

Aplanospore or Akinete?

In the original account of *G. polyrhiza* Bornet and Flahault describe what they termed aplanospores. These were about 4 μ in diameter, the size of the small zoospore, and their development into large irregular cells which persisted for a long time without change is practically the same as described above for the resting spore. In fact there can be no doubt that the resultant body from the so-called aplanospore of Bornet and Flahault and that described by me as originating from a zoospore is the same thing. It will be recalled, however, that Bornet and Flahault never saw the discharge of either the zoospores or aplanospores, nor did they observe the germination of the small zoospores described by them. It would therefore seem possible that the so-called aplanospores of Bornet and Flahault were nothing but zoospores which had come to rest. Plate 13, fig. 5, shows a few zoospores which, failing to escape, rounded up and began the development of resting spores. These might easily be mistaken for aplanospores. Certainly in all the material of *G. lignicola* which was examined under much more favorable circumstances than was possible by Bornet and Flahault, at no time was there the slightest indication of the production of an aplanosporangium or anything which might be regarded as a true aplanospore. Furthermore, in the description of no other species of *Gomontia* is there given any evidence of the existence of aplanospores, their presence in the genus being based apparently entirely upon the original account of Bornet and Flahault. It seems very doubtful, therefore, that aplanospores exist in any species of *Gomontia*. The authority for West's[1] statement that akinetes are found in *Gomontia* is apparently based upon Chodat's account of *G. codiolifera;* at least he cites a figure from Chodat as an example of akinete formation. Chodat called this figure an example of "états Codiolum deviennent finalement des zoosporanges." It is without doubt a resting spore developed from a zoospore, and, so far as known, there is no justification for considering akinetes as existing in any species of *Gomontia.*

Gomontia lignicola Moore, n. sp.

Type: in Mo. Bot. Gard. Herb.

Filaments usually simple, rarely branched, not radiating from a common center; a single parietal chloroplast much reticulated, usually most abundant in apical cell; numerous pyrenoids; 1–6 nuclei; mature cells 25–45 μ in width, 100–200 μ in length; zoosporangia typical for the genus; zoospores

[1] Algae 1 : 305. 1916.

biciliate, 10–12×12–15 μ; germination either direct or by producing large irregular-shaped resting spores which germinate direct, producing one or more vegetative filaments; no aplanospores or akinetes observed.

Growing within tissue of wood (yellow pine) submerged in fresh water; to be regarded as at least partially aërial in habit. Fresh pond on Nashawena, Elizabeth Islands, Massachusetts.

Explanation of Plate

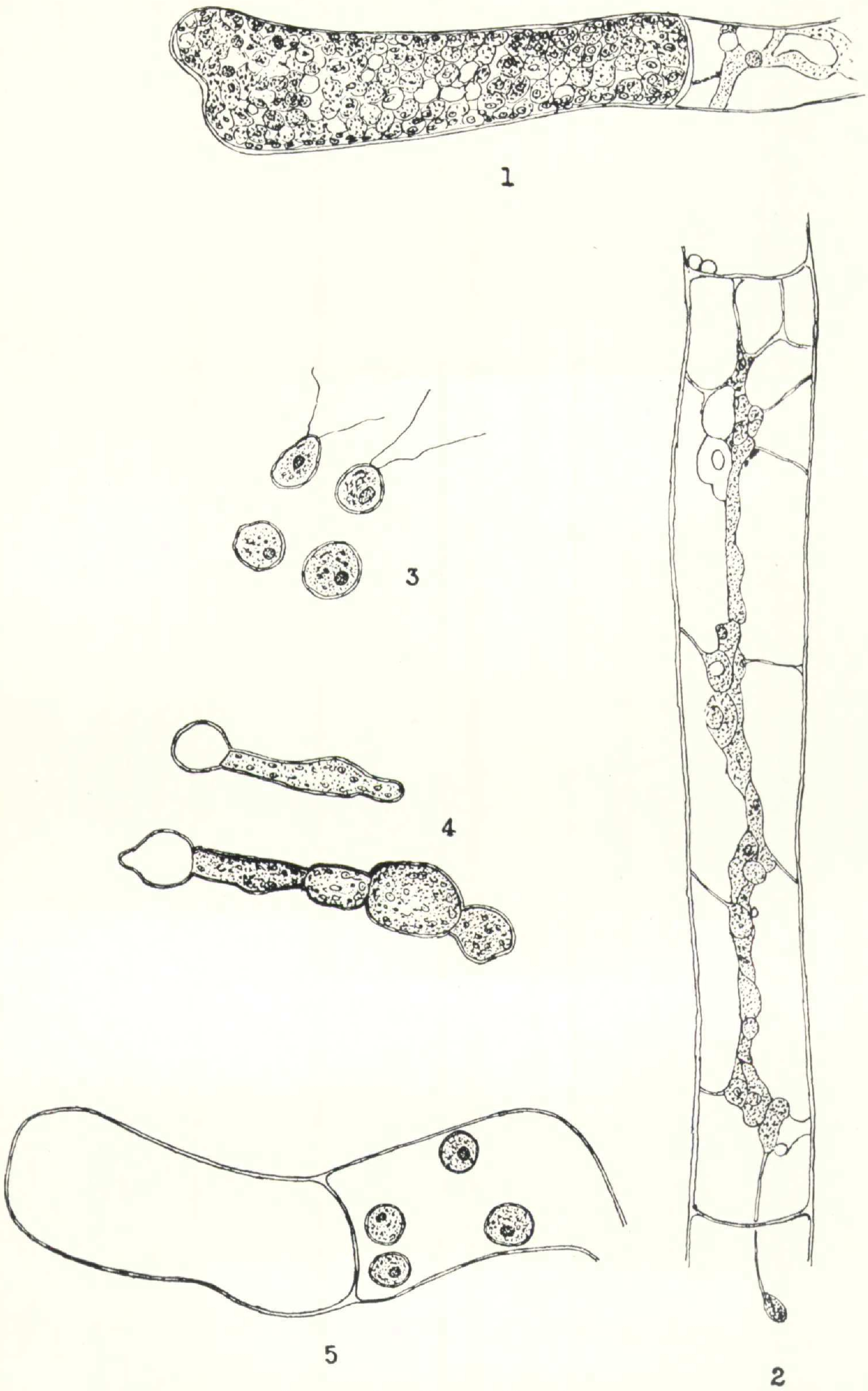
PLATE 13

All figures are reproduced from camera drawings × 580.

Figs. 1 and 2. Terminal and contiguous vegetative cells, showing difference in chloroplast, also protoplasmic connection.

Figs. 3 and 4. Zoospores and direct germination.

Fig. 5. Zoospores which have come to rest within sporangium.



MOORE—GOMONTIA LIGNICOLA

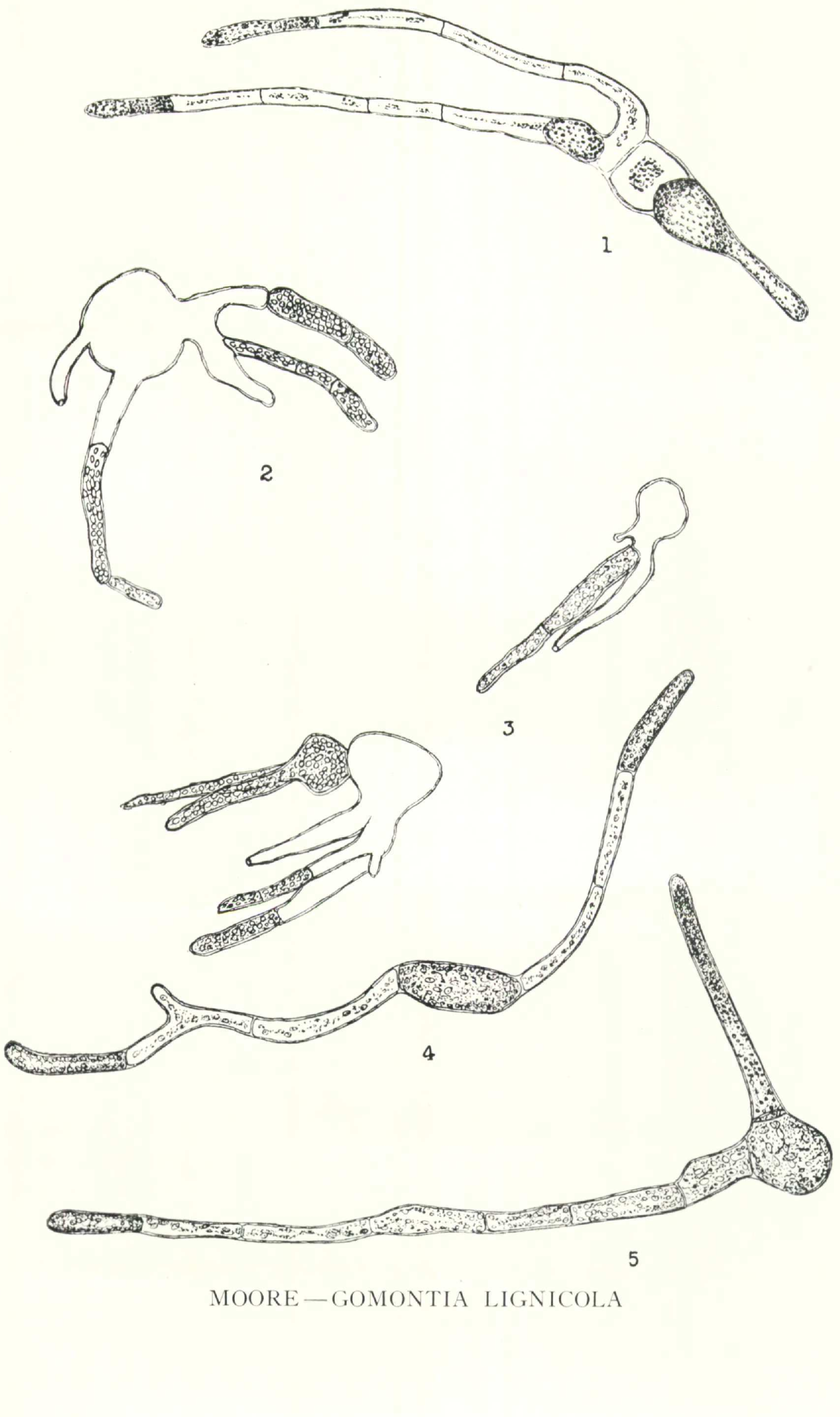
Explanation of Plate

PLATE 14

All figures are reproduced from camera drawings × 100.

Figs. 1, 4, and 5. Germinating resting spores, showing vegetative filaments.

Figs. 2 and 3. Sporangia partially empty.



MOORE—GOMONTIA LIGNICOLA

Explanation of Plate

PLATE 15

All figures are reproduced from camera drawings × 580.

Various stages in development of resting spores from zoospores, as well as a few examples of mature resting spores.